Radioimmunotherapy of Patients with Small-Volume Tumors Using Iodine-131-Labeled Anti-CEA Monoclonal Antibody NP-4 F(ab')$_2$

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The clinical feasibility of radioimmunotherapy (RAIT) was assessed in patients with metastatic, carcinoembryonic antigen (CEA)-producing cancers who had minimal residual or small-volume disease (tumor lesions ≤3 cm in diameter). **Methods:** Thirteen cancer patients (8 colorectal, 3 lung, 1 pancreatic and 1 medullary thyroid cancer) received RAIT with ^{131}I-NP-4 F(ab')$_2$ anti-CEA antibody. The radioactive dose given was based on a prescribed radiation dose to the red marrow. Ten of the 13 patients received initial therapeutic doses delivering 150–450 cGy to the red marrow (70–296 mCi) and six patients had more than one therapy infusion. **Results:** Targeting of all known tumor lesions 70.5 cm in diameter was possible in nine patients and at least one tumor lesion was evident in all patients. Disease stabilization ranging from 3.5 to 7 mo was seen in 6 of the 13 patients who previously had clear evidence of progressive disease. Four of the six patients with disease stabilization received the presumed maximum tolerated dose of 450 cGy to the red marrow. Red marrow suppression was the only observed toxicity and there was a good correlation between the red marrow dose and myelotoxicity. Red marrow doses ≤250 cGy resulted in ≤grade 2 myelotoxicity and a red marrow dose of 450 cGy resulted in reversible grade 3 or 4 myelotoxicity in 3 of 6 patients. Human anti-mouse antibodies (HAMA) developed in all but one of the six patients who received multiple therapeutic infusions of the antibody. **Conclusion:** RAIT of patients with small-volume disease is feasible and these patients should be considered for future dose-intensification trials because of their generally poor prognosis.

Key Words: carcinoembryonic antigen; monoclonal antibodies; radioimmunotherapy; small-volume disease

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Encouraging results have been obtained in the treatment of chemotherapy-resistant Hodgkin's and non-Hodgkin's lymphomas with radiolabeled monoclonal antibodies (MAbs) (*1–6*). One of the most important factors that led to successful radioimmunotherapy (RAIT) of these malignancies is their inherent radiosensitivity compared with a wide range of solid tumors (*1–8*). This factor becomes more apparent when one correlates the radiation doses delivered to these different tumors with the responses achieved. While partial and even complete remissions have been reported in a substantial percentage of lymphoma patients with tumor doses <500 cGy (*1–5*), objective antitumor responses have been observed infrequently in the RAIT of solid tumors (*9–16*), including colorectal cancer patients, even with tumor doses as high as 3300 cGy (*16*).

If the experience with radioiodine therapy of differentiated thyroid cancer can be applied to the RAIT of most solid tumors, notwithstanding the differences existing in the radiation sensitivity of various solid tumor types, then tumor radiation doses of at least 3500 cGy may be necessary to obtain objective responses (*15*). Achieving such tumor doses, however, has been difficult in most clinical RAIT trials. This is primarily due to the low average percent injected dose accreted in tumor, which is usually in the range of 0.001% and 0.01% per gram of tumor (*16,17*). Perhaps one of the most important reasons for these low tumor doses with RAIT is the large size of most tumors treated. Several investigators, including our group, have reported an inverse relationship between size and tumor uptake in humans (*18*) and in animal models (*19–22*). In addition, the most impressive antitumor effects in the RAIT of solid tumors are seen in patients with minimal residual or small-volume disease (*9,10*). These results have prompted us to initiate an exploratory study to assess the clinical feasibility of RAIT in this group of patients.

In this article, we describe the results of this RAIT study with single or multiple infusions of ^{131}I-labeled anti-CEA antibody F(ab')$_2$ in 13 patients with metastatic CEA-producing cancers, in whom the size of tumor lesions was restricted to ≤3 cm in diameter. The results of this initial study are encouraging, especially in patients who received high-single or multiple doses of radioactivity.

MATERIALS AND METHODS

Patients

Patients with histologically-proven, CEA-producing cancers (including colorectal, pancreas, lung, breast, ovary, endometrial and medullary thyroid) of ≤3 cm in diameter were eligible for this study. To enter the therapy studies, the patients had to be at least 4 wk beyond any major surgery, radiation or chemotherapy, and had to have recovered from any prior treatment-induced toxicity. The patients had a performance status of ≥70 on the Karnofsky scale (ECOG 0–2) and a minimal life expectancy of 3 mo; no severe anorexia, nausea or vomiting; normal hepatic and renal function; WBC ≥3000/mm^3 or a granulocyte count ≥1500/mm^3 and a platelet count ≥100,000. Subjects were excluded from treatment if they were pregnant or if they have had extensive irradiation to more than 25% of their red marrow within 1 yr of treatment. The patients were mentally responsible and gave informed consent. All protocols were approved by the governing Institutional Review Board.

A total of 13 patients (6 men, 7 women; age range 40–80 yr) received single or multiple therapeutic doses of ^{131}I-NP-4 F(ab')$_2$. Eight patients had colorectal, three had lung, one had pancreatic and one had medullary thyroid cancer. The mean plasma CEA level was 120 ng/ml (s.d. = 200 ng/ml), but plasma CEA was ≤100 ng/ml (range 3.0–100.0 ng/ml) in 11 of 13 patients studied. Only two patients had CEA levels of 398 and 683 ng/ml, respectively. Table 1 lists the patient data, antibody infusion and the site of disease.

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TABLE 1
Antibody Infusions and Sites of Disease

Patient no.	Sex	Age	Cancer type	(Inj.)	Date*	Antibody infusion	Sites of disease†
1036	F	59	Colon	1	0	10.2 mCi (0.7 mg)	Lungs
				2	2	238.0 mCi (25.2 mg)	
				3	10	200.2 mCi (13.6 mg)§	
				4	22	151.9 mCi (12.7 mg)§	
1063	F	79	Colon	1	0	7.9 mCi (0.7 mg)	Liver bone
				2	4	103.4 (11.5)§	
1124	M	43	Colon	1	0	68.5 (8.2 mg)	Lungs, bone
1125	F	54	Colon	1	0	67.5 (8.2 mg)	Liver
				2	4	228.4 (26.8 mg)§	
1156	M	65	Colon	1	0	76.1 (6.3 mg)	Local recurrence, liver,
					6	254.0 (18.5 mg)	periaortic lymph nodes
1217	F	49	Colon	1	0	72.0 (5.6 mg)	Lungs
					8	164.7 (16.3 mg)	
					18	121.4 (241.4 mg)‡	
					38	240.6 (240.6 mg)‡	
991	F	62	Rectal	1	0	9.8 mCi (1.1 mg)	Lungs
				2	6	177.3 mCi (17.4 mg)	
				3	15	120.0 mCi (17.0 mg)‡	
1186	M	66	Rectal	1	0	63.2 (7.8 mg)	Lungs
				2	8	229.5 (25.1 mg)‡	
775	F	69	Lung	1	0	70.0 mCi (14 mg)	Bone, lungs,
				2	8	68.1 mCi (6.5 mg)	adrenal glands
				3	17	78.7 mCi (5.6 mg)	
				4	53	88.6 mCi (10.3 mg)	
1047	F	54	Lung	1	0	10.7 mCi (1.0 mg)	Lung, liver,
				2	8	203.0 mCi (20.4 mg)	brain
1142	M	77	Lung	1	0	80.8 (9.4 mg)	Primary tumor, lungs, mediastinal and hilar nodes, right adrenal
1014	F	68	Pancreatic	1	0	12.0 mCi (1.2 mg)	Lungs, primary
				2	10	151.4 mCi (14.0 mg)	tumor, portal
				3	19	64.4 mCi (6.0 mg)‡	and mesenteric
				4	40	65.0 mCi (5.9 mg)‡	lymph nodes
1318	M	40	MTC	1	0	110.9 mCi (13.0 mg)	Cervical lymph
				2	32	243.8 mCi (25.5 mg)‡	nodes

*0 = indicates the date of the first infusion, the numbers indicate the weeks thereafter.
†Site of disease by CT, MRI, bone scan, radiograph, ultrasound or surgery at the time of admission.
‡HAMA level <500 ng/ml.
§HAMA level >500 ng/ml (dose injected in parentheses).

Radiolabeled Antibody Preparation

The murine IgG_1 NP-4 MAb is directed against the Class-III, CEA-specific epitope according to the classification of Primus et al. (*23*). The $F(ab')_2$ fragment was prepared by pepsin digestion. All preparations used passed general safety testing. NP-4 $F(ab')_2$ was labeled with ^{131}I-Na by the iodogen method to a specific activity of 12–16 mCi/mg, as previously reported (*24*). More than 70% binding to a CEA immunoadsorbent was found for the radiolabeled antibody. Less than 2% unbound isotope and <7% aggregation were demonstrated by high-performance liquid chromatography (HPLC) for all agents.

Antibody Infusions

The radioactivity given was based on a prescribed red marrow dose and was determined by a pretherapy tracer study with 10–15 mCi or by a dosimetry infusion with 40 mCi/m^2 of ^{131}I-NP-4 $F(ab')_2$, originally given to obtain high quality images at later time points. In cases where a dosimetry infusion was given prior to RAIT, the sum of the red marrow dose of both infusions was considered if both infusions were given within an 8-wk interval.

The maximum tolerated dose (MTD) with ^{131}I-NP-4 $F(ab')_2$ (i.e., the dose that results in only transient grade 3 or 4 myelotoxicity) was previously determined by our group as 450 cGy to the red marrow (*25*). In this study, however, not all patients received the MTD. Our intent was to also obtain an initial experience in the treatment of small-volume or minimal residual disease with doses that were lower than the MTD (i.e., doses that result in ≤grade 2) myelotoxicity. Therefore, three patients received the dosimetry infusions only (40 mCi/m^2) delivering 100 to 150 cGy to the red marrow, one patient received a red marrow dose of 250 cGy, and six patients received the MTD of 450 cGy. Two patients were scheduled to receive the MTD of 450 cGy, but in one patient with lung cancer (Patient 1142), the dosimetry infusion cleared rapidly, resulting in a red marrow dose of only 55 cGy. The rapid clearance in this patient was probably related to pre-existing HAMA; thus, the patient did not receive further treatment. In the other patient (Patient 1125), high levels of HAMA developed after the initial dosimetry study, resulting in a very rapid clearance of the therapy infusion and a total red marrow dose of only 191 cGy. In Patient 1063, high levels of HAMA developed after the initial tracer study, resulting in a red marrow dose of ~55 cGy in her therapy infusion. Of the 13 patients, six received more than one therapy infusion (mean = 2.8, range: 2–4 infusions). All injections were given intravenously, proceeding slowly over the first 5 min, and then at a more rapid rate to complete the infusion within 15–30 min. No

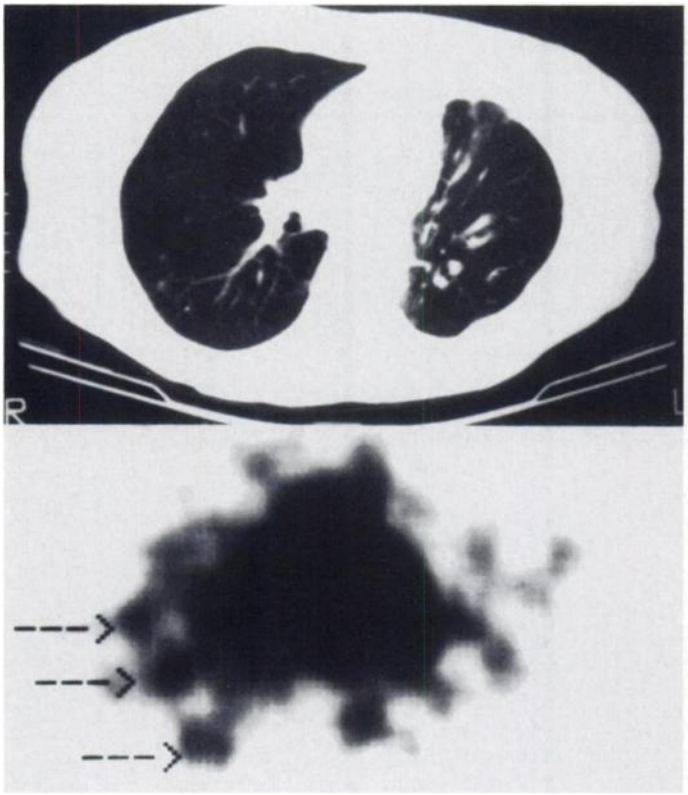

FIGURE 1. CT of the chest demonstrating three small pulmonary nodules (arrows) in the right lung. A transverse SPECT section corresponding to the CT shows targeting of these nodules with ^{131}I NP-4 F(ab')$_2$. Note that the nodules appear larger on SPECT (and apparently less intense) due to the limited spatial resolution of the gamma camera.

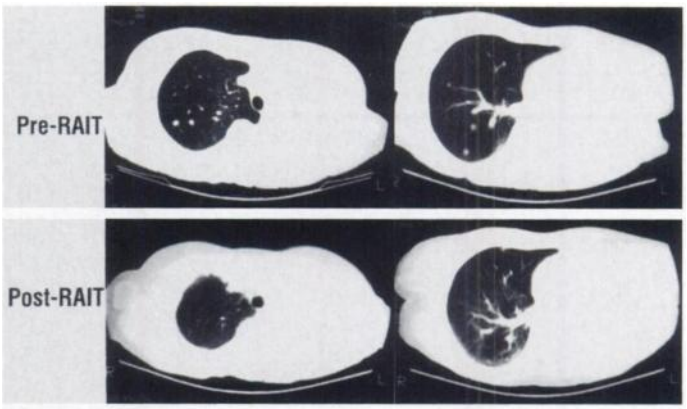

FIGURE 2. CT scans of the chest 2 days before (top) and approximately 2 mo after a second RAIT (bottom) with 68.1 mCi ^{131}I NP-4 F(ab')$_2$ in Patient 775. Scans are taken at the level of the aortic arch and right lower lobe artery show the disappearance of two small nodules in the right upper lobe and two other nodules in the right lower lobe. No significant change was seen in at least two other nodules.

adverse experiences were observed with any of the injections, including patients with HAMA. All patients were premedicated with Lugol's solution (five drops orally, three times per day) and potassium perchlorate (200 mg orally, twice per day) to decrease thyroid and gastric uptake of radioiodine, respectively.

HAMA Monitoring and Follow-up Studies

A baseline plasma HAMA titer was determined in all patients, with follow-up at 1, 2, 4, 8 and 12 wk using a HAMA titer assay (*26*) or, more recently, the ImmuSTRIP™ HAMA IgG assay (Immunomedics, Inc., Morris Plains, NJ). Normal values for the ImmunSTRIP™ assay are <74 ng/ml. Correlative radiological studies, such as CT, were performed within 4 wk, usually within 1–2 wk, prior to antibody imaging or treatment, with follow-up CT studies performed at a minimum of 1 and 3 mo. Circulating CEA was measured on the day of treatment and at 1–3-mo intervals for 1 yr or more thereafter. CEA was determined in heat-extracted plasma samples to eliminate interference with HAMA (*26*). Other assays were performed by registered clinical laboratories.

Pharmacokinetic Analysis

Blood clearance rates were determined by counting samples of whole blood at various time points after the end of the infusion. Three to five blood samples were taken over the first 24 hr, and then daily sampling was performed over the next 2–6 days. Curve-fitting programs were used to generate both monoexponential or biexponential clearance curves, as reported previously (*3*). Total-body clearance rates were determined by: (a) whole-body external scintigraphy taken at three separate times, (b) daily measurements with a rate meter at 1 m from the patient or (c) urine collection. The term $T^{1/2}$ used in this report indicates the time (in hours) taken to clear 50% of the initial radioactivity from the blood or body.

Imaging and Dosimetry

Planar images (500 kcts per view) consisting of anterior and posterior scans of the head, chest, abdomen and pelvis were obtained using DS-X or DS-7 Sopha cameras (Sopha Medical Systems, Columbia, MD) equipped with a high-energy collimator. Images were taken at 4 hr and then daily for up to 1 wk postinfusion of a diagnostic dose of the antibodies using a 128 × 128 matrix. Post-therapy imaging was initiated when the level of activity fell below 5 mR/hr at 1 m, usually 3–5 days postantibody infusion. SPECT studies (64 × 64 matrix-size) of the chest, abdomen and pelvis were obtained on at least one occasion 24 hr or later postinfusion. SPECT was used to detect the tumor site by improved contrast resolution. An activity quantification technique for the gamma camera was used for the dosimetric calculations as described previously (*18*). Tumor volumes were measured by CT and standard organ weights given by MIRD were used (*27*). The organ and tumor time-activity data were then fit to either an exponential function by a nonlinear, least-squares, curve-fitting routine, or by a trapezoidal modeling method, and then integrated to obtain the cumulated activity. The cumulated activity in the red marrow was calculated from the blood by multiplying this concentration by 1500, the weight in grams of the marrow in an average adult (*28*). The mean dose in cGy (rad) to the various target organs, with the exception of the tumors, was then obtained according to the MIRD schema with correction for the remainder of the body activity (*27,29*). The mean dose in cGy to the tumors was obtained by the previously method reported (*18*).

Toxicity and Tumor Response

Toxicity was graded according to the Radiation Therapy Oncology Group (RTOG) criteria. All patients given therapeutic doses of ^{131}I-MAbs were followed for toxicity by monitoring complete peripheral blood cell counts weekly. Renal and hepatic functions were assayed 7 and 28 days post-therapy. Tumor responses were assessed 1–3 mo after treatment and every 3 mo thereafter, up to 1 yr. If disease progression occurred after 3 mo, no further follow-up was attempted. In addition to physical exams, chest radiograph, CT and MRI were used to assess therapeutic response. Plasma CEA level was assessed at 1 to 3 mo, up to 1 yr post-therapy. Reduction in CEA level that was >25% for at least 1 mo was considered to be an indication of an anti-tumor effect. Complete remission was defined as the complete disappearance of all detectable disease for a minimum of 4 wk, a partial response as a reduction of at least 50% in the sum of the products of the longest perpendicular diameters of all measurable lesions for a minimum of 4 wk, and

TABLE 2
Therapeutic Results in Initial Assessable Patients

Patient no.	No. of treatments	Initial red marrow dose (cGy)	Anti-tumor effects (duration)
1142	1	55	Progression
1063	1	55	Stable disease (1 mo)
1124	1	108	Progression
775	4	150*	Stabilization of disease (7 mo) (Regression of small pulmonary nodules and stable adrenal and bony metastases)
1125	2	194†	Stabilization of disease (5 mo) [25% reduction in left cervical and stable right cervical adenopathy. Stable CEA and calcitonin (11 mo)]
1318	1	191	Progression
1047	1	250	Progression
991	2	450	Stabilization of disease (3.5 mo)
1014	3	450	Stabilization of disease (5 mo)
1036	3	450	Stabilization of disease (6.5 mo)
1217	3	450	Stabilization of disease (7 mo)
1156	1	450	Progression
1186	1	450	Progression

*Patient received two additional RAIT cycles in 2-mo intervals, delivering a total red marrow dose of 501 cGy.

†Red marrow dose after the second RAIT, 10 mo after the first RAIT, delivering a total dose of 150 cGy to the marrow.

disease progression as an increase of at least 25% in the diameter or the appearance of new lesions. Minor responses were considered when the reduction in disease was between 25% and 50%.

RESULTS

Pharmacokinetics

Pharmacokinetic data were available in all 13 patients given the dosimetric or therapeutic doses of ^{131}I-labeled NP-4 F(ab′)$_2$. HPLC performed at 1 hr has shown that <15% of the antibody was Fab′, except in one patient with 24% Fab′. The 24-hr HPLC, however, showed that three patients had Fab′ fractions >15% (range 17–42%). High molecular weight complexes >20% were seen in only one patient (Patient 1063), in whom the HPLC revealed 59% and 90% antibody complexation in plasma at 1 and 24 hr, respectively. This patient, whose pretreatment CEA was 681 ng/ml, received a tracer dose of 7.9 mCi (0.7 mg) of NP-4 F(ab′)$_2$ 5 wk prior to the therapeutic infusion. A HAMA assay performed at the time of the therapy infusion revealed an elevated level of 745 ng/ml. Accordingly, the antibody cleared in this patient with a biological half-life of only 2.5 hr. In contrast, only 13% of the injected tracer dose was complexed and the antibody cleared with a biological half-life of 31.5 hr. The average $T_{1/2}$ in blood and total body for NP-4 F(ab′)$_2$ in the 12 patients with a HAMA level <100 ng/ml was 15.3 hr ± 4.7 hr and 49.5 hr ± 13.1 hr, respectively. As expected, HAMA developing after repeated antibody infusions had a profound effect on antibody clearance, resulting in a 1.3- to 12-fold decrease in the residence time of NP-4 F(ab′)$_2$ in the blood, depending on the plasma HAMA level. Complexation with HAMA also altered the distribution of the radioactivity, with enhanced uptake in the liver, spleen and bone marrow, as well as to the normal thyroid. Tumor uptake was adversely affected, by qualitative assessment, in all patients who developed HAMA. In patients with high levels of HAMA (i.e., >500 ng/ml), there was only faint tumor uptake. In other patients with HAMA levels <500 ng/ml, there was a mild-to-moderate decrease in tumor uptake compared with the baseline. Quantitative assessment of the effect of HAMA was possible in one patient (Patient 1125) with a HAMA level of 12,510 ng/ml, who had a 72% reduction of tumor uptake (in % ID/g) compared with the baseline.

TABLE 3
Myelotoxicity after Initial Therapeutic Infusion of Iodine-131-NP-4 F(ab′)$_2$

Patient no.	Radiation dose to red marrow (cGy)	Myelotoxicity grade	TTR
1063	55	0	na
1142	55	0	na
1124	108	0	na
775	150	2 WBC, 1 PLT	21
1318	150	0	na
1125	191	0	na
1047	250	2 PLT, 1 RBC	27
991	450	3 WBC, 2 PLT	35
1014	450	4 WBC, 4 PLT	36
1036	450	2 WBC, 2 PLT	17
1156	450	1 WBC	nd
1186	450	4 PLT, 3 WBC, 1 RBC	41
1217	450	1 WBC, 1 PLT, 1 RBC	17

WBC = white blood cells; PLT = platelets; RBC = red blood cells, TTR = time to full recovery from the nadir of the most severe myelotoxicity (in days); na = not available; nd = not determined.

HAMA

Of the five patients who received a tracer antibody dose (<1 mg) of ^{131}I-NP-4 F(ab′)$_2$ within 2–8 wk prior to the administration of therapy infusion, only one patient developed an elevated HAMA. Four of eight patients who received a higher initial dose of 5–10 mg of the antibody, however, developed HAMA. All five patients developed HAMA more than 3 wk after the infusion. All but one of six patients who received two or more therapy infusions developed HAMA after their second therapy infusion, resulting in more rapid clearance of the subsequent antibody infusions.

Targeting

Targeting of at least one known disease site was seen in all patients who received the therapeutic doses of ^{131}I-NP-4 F(ab′)$_2$. All disease sites 0.5 cm in diameter were visualized in 9 of 13 patients studied. Confirmed tumor lesions, however, could not be disclosed by external imaging in four patients. In Patient 1124 with colonic cancer, there was no definite visualization of lung lesions of varying sizes (0.2–1.0 cm), despite

TABLE 4
Whole Body and Organ Dosimetry (in cGy/mCi) for Iodine-131-NP-4 F(ab′)$_2$

Whole body	Red marrow	Kidneys	Liver	Lung	Spleen
0.5 ± 0.1 (n = 8)	1.8 ± 0.4 (n = 8)	3.3 ± 1.9 (n = 7)	1.6 ± 0.7 (n = 8)	2.1 ± 0.6 (n = 8)	4.4 ± 2.7 (n = 7)

targeting of known bone metastases. Patient 1014 with pancreatic cancer also failed to visualize multiple 0.2–0.6 cm lung nodules, but the disease was targeted in the primary tumor and in portal lymph nodes. Patient 1047 with lung cancer showed targeting of 0.5 to 3.0 cm lesions in the liver, but no definite visualization of the known 0.2–1 cm lung lesions or 1.8 cm brain lesion. Patient 1156 showed targeting of the periaortic lymph node metastases, but failed to visualize a local recurrence in the colon. This was also negative on CT but positive by colonoscopy and at surgery. In many instances, imaging of small lesions (i.e., <1 cm) was only possible with SPECT. Figure 1 shows an example of targeting of small lung lesions (confirmed by CT) using SPECT in a patient with colonic cancer. This patient was not included in this study, since he also had liver and bony metastases > 3 cm in size.

Initial Therapeutic Results

No major tumor responses were seen in this study. Disease stabilization >1 mo (range, 3.5–7 mo, median: 5 mo) was seen in six patients who previously had evidence of clearly progressive disease by tumor markers and/or radiological studies. Two of these patients showed regression of individual tumor lesions seen on a follow-up CT scan or by physical exam; however, the overall reduction in tumor burden was <50% and, therefore, could not be considered a partial remission. The disease progressed in six patients within 1–3 mo after RAIT.

Four of the six patients who showed disease stabilization after RAIT received the maximum tolerated dose (450 cGy to the red marrow) given as a single infusion (three patients) or in two infusions administered 6 wk apart (one patient). One additional patient received three therapeutic infusions of ^{131}I-NP-4 F(ab′)$_2$ at 2-mo intervals without developing HAMA. The cumulative red marrow dose in this patient was 501 cGy. The last patient showed stable disease after a second therapy infusion delivering 194 cGy to the red marrow. The second infusion was given 8 mo after the initial therapy, which delivered 150 cGy to the red marrow. In contrast, only two of the six patients with disease progression received the MTD of 450 cGy. Moreover, three of the six patients had either a single low dose of ^{131}I-NP-4 F(ab′)$_2$ delivering <110 cGy to the red marrow (two patients) or developed a high level of HAMA after the initial tracer study, resulting in a rapid clearance of the second antibody infusion and a red marrow dose of only 55 cGy (one patient).

The course of disease after single or multiple therapeutic infusions of ^{131}I-NP-4 F(ab′)$_2$ is illustrated in Patients 775 and 1318. Patient 775 had poorly differentiated adenocarcinoma of the lung. Baseline CT scans of the chest, abdomen and pelvis performed at the time of her referral demonstrated multiple lung nodules (all <1.5 cm), a right adrenal mass and multiple bony lesions in the lower lumbar spine and pelvis, which were also seen on a baseline bone scan. The right adrenal mass was new compared to a CT scan obtained 2 mo before her entry into the RAIT trial. In addition, there was clear evidence of disease progression in the lung on two consecutive CT scans of the chest obtained within 4 mo of therapy. Her CEA also rose from 34 ng/ml to 82 ng/ml over a 3-mo period prior to entering the RAIT trial. The patient first received 70 mCi, followed 2 mo later by 68 mCi of ^{131}I-NP-4 F(ab′)$_2$. A chest CT scan performed 2 mo after the second therapy showed regression of four small pulmonary nodules, without significant change in the other nodules or the appearance of new lesions (Fig. 2). Overall, the disease was found to be stable in the lung, right adrenal and bone. The patient's plasma CEA level also remained stable over the course of the last 4 mo, with values between 50 and 77 ng/ml. Two months after the second RAIT, the patient received a third infusion of 79 mCi ^{131}I-NP-4 F(ab′)$_2$. The disease remained stable until 3 mo after this third treatment, when CT scans demonstrated progression. Fourteen months after her initial treatment, the patient's CEA nearly doubled, and because there were no other therapeutic options, a fourth treatment of 88.6 mCi ^{131}I-NP-4 F(ab′)$_2$ was given. The patient developed HAMA during the course of treatment, resulting in a rapid clearance of the radiolabeled MAb. She was therefore not eligible to receive further treatment. Her disease progressed further and she died 8 mo later. In this patient, the disease was considered to be stable for 7 mo during the initial three cycles of RAIT. Only moderate myelotoxicity (≤grade 2 leukopenia and thrombocytopenia) developed during the course of treatment.

Patient 1318 had progressive medullary thyroid cancer by tumor markers and CT. He first received 111 mCi of ^{131}I-NP-4 F(ab′)$_2$. The 1-mo follow-up CT scan showed stable disease in the neck. Due to persistent extensive cervical adenopathy, however, the patient underwent bilateral lymph node dissection. Two months after the operation, there again was evidence of progression in the neck and the disease was judged to be inoperable. The patient then received his second RAIT with 243.8 mCi of ^{131}I-NP-4 F(ab′)$_2$ 8 mo after the first treatment. Despite a HAMA titer of approximately 500 ng/ml, targeting of cervical adenopathy was seen. Two months after RAIT, a minor reduction (25%) of his left cervical adenopathy was seen by CT, and disease stabilization was noted on the right side. His CEA and calcitonin also remained stable at ~25 ng/ml and 25,000 pg/ml, respectively. The disease remained stable for approximately 5 mo, when progression was noted by physical exam and CT. The patient did not develop any myelotoxicity after the first treatment, and only a grade 1 leukopenia after the second treatment at a red marrow dose of 194 cGy. The patient recovered from leukopenia in only 7 days.

Toxicity

There was relatively good correlation between the red marrow dose calculated by blood and the myelotoxicity seen (Table 3). Red marrow doses ≤250 cGy resulted in ≤grade 2 myelotoxicity. Of the 13 patients treated, only three had a transient grade 3 or 4 myelotoxicity after their initial therapy infusion. All of these patients received the MTD of 450 cGy to the red marrow as a single infusion (two patients), or in two infusions given 8-wk apart (one patient).

Organ and Tumor Dosimetry

Table 4 shows the mean organ doses (in cGy/mCi) obtained with ^{131}I-NP-4 F(ab′)$_2$. As stated earlier, tumor targeting of lesions <1 cm was only possible using SPECT (see Fig. 1) and, thus, it was not possible to calculate the dose for those tumor lesions not seen on planar scans. Doses, however, were calculated for four tumor lesions of 1.8 to 2.4 cm in size. CT was then used to calculate the tumor mass based on the assumption of a sphere. The estimated tumor masses were between 3 and 7 g. The estimated radiation doses delivered to 3 lesions (lymph node metastases) in one patient (Patient 1318) were 511, 578 and 1100 cGy. The dose delivered to a fourth lesion (liver metastasis) in one patient was 6476 cGy. Unfortunately, this patient developed brain metastases, received external-beam whole-brain radiation, and was then lost to follow-up. The mean percent injected dose per gram tumor (%ID/g) at the time of maximum uptake was 0.04% (range 0.004–0.12%). The mean biological half-life in tumor was 197 hr (range 30–437 hr), and the estimated mean tumor dose was 12.9 cGy/mCi (range 4.6–31.9 cGy/mCi).

DISCUSSION

This study demonstrates the clinical feasibility of RAIT in cancer patients with small-volume or minimal residual disease. Our hypothesis was that these patients may be more suitable for RAIT due to the possibly higher uptake of antibody in small tumor lesions (*18–22*). Two types of patients with small-volume disease were referred for RAIT. One group included those who received conventional chemotherapy at the time of modest elevations of tumor markers, sometimes even without radiological evidence of disease, but continued to progress and were therefore considered to be chemotherapy-resistant. The other group included patients who had biochemical or radiological evidence of disease relapse after primary surgery. These patients were then referred at the time of clear evidence of disease progression before they received standard chemotherapy.

Consistent with the relatively small tumor burden in most patients (due to the small size of tumor lesions), the CEA plasma level was only moderately elevated. The mean value was 120 ng/ml, with 11 of 13 patients having CEA levels of <100 ng/ml. Rosenblum et al. (*30*) have previously reported a negative correlation between elevated CEA levels and plasma clearance in colorectal cancer patients given NP-4 $F(ab')_2$. This is most likely related to the formation of CEA-antibody complexes that are rapidly cleared and metabolized in the liver. In this series, the moderate elevation of CEA, in addition to the moderate affinity of NP-4, were probably responsible for the very low complexation of antibody with the patient's plasma CEA and, therefore, relatively normal antibody blood clearance.

Tumor imaging of at least one tumor lesion was possible in all patients when SPECT was used in addition to planar imaging. It is important, however, to note that not all tumor lesions could be visualized in 4 of 13 patients, even with the use of SPECT. While we cannot exclude that ex vivo counting may be able to demonstrate increased antibody accumulation in these lesions, poor visualization of lesions >0.5 cm may be considered indicative of relatively poor tumor targeting and may explain the lack of response to radiation in some patients.

Since we currently utilize planar imaging for obtaining organ and tumor dosimetry, it was not possible to calculate the radiation dose to small tumors (<1 cm) not seen on planar scans, as was the case for most lesions in this study. The mean tumor dose for the four lesions of 1.8 to 2.4 cm in diameter (3 to 7 g) was 12.9 cGy/mCi, and 0.04 percent of the injected dose was found per gram of tumor. While this value is several fold higher than the usually reported 0.001 to 0.01% ID/g, higher values may be expected for <1 cm (0.5 g) lesions, as has been reported by other investigators (*31*). We are currently evaluating the use of SPECT for computing the dosimetry of these small lesions, so that a reliable estimate of radiation dose to these tumors may be possible in future studies.

It is very important to note that minimal residual or small-volume disease is not synonymous with stable disease. In fact, the vast majority of patients in this series had clear evidence of disease progression by CT scans and by tumor markers prior to their referral for RAIT. Moreover, the median survival of the 11 patients who died of their cancer was only 8 mo from the time of study entry, clearly indicating their dismal prognosis. It is therefore encouraging that disease stabilization ranging from 3.5 to 7 mo was noted in six of these patients. These results are particularly encouraging, since the maximum tolerated dose was given only to six patients, four of whom showed disease stabilization. The fact that major tumor responses were not seen and that disease stabilization was observed in patients who received relatively high cumulative doses argues for the need for dose intensification. One strategy for achieving this goal is repeated administration of the MTD without autologous bone marrow or stem cell support. Unfortunately, this strategy is rarely successful when murine antibodies are used, because of the development of human anti-mouse antibodies. In this series, and despite the lower immunogenicity of $F(ab')_2$ fragment compared with the whole IgG, administration of more than two therapy infusions without HAMA development was only possible in one patient. Moreover, HAMA developed as early as 3 wk after the MAb infusion in 4 of 8 patients who received 5–10 mg of protein in their initial dose, thus impeding the administration of a second dose. When the MTD is given, 8 to 12 wk may be needed for full recovery of the red marrow before a second therapeutic dose can be given. Therefore, there is a high likelihood that HAMA will develop in these patients. In addition, higher protein doses are expected to be given at the MTD, which will result in even greater probability of HAMA development. With the advent of human or humanized antibodies, however, the repeated administration of the MTD should become feasible (*32*). Our preliminary data indicate that a red marrow dose of 450 cGy results in only transient grade 3 or 4 toxicity. In fact, the duration of grade 3 toxicity was 14 days in one patient, and that of grade 4 was 2 and 5 days in two patients, respectively. Even though a longer period is needed for full recovery of the red marrow, the transient nature of myelotoxicity in these patients may allow the administration of multiple doses without the need for red marrow or stem cell support.

Another strategy to increase the therapeutic efficacy of RAIT in this group of patients is the administration of high doses of radiolabeled antibodies combined with autologous red marrow or stem cell support (*5,14*). This regimen is justified considering the poor prognosis of patients with metastatic colorectal, pancreatic, lung, breast, and ovarian cancer, even if they have minimal residual disease. In fact, these patients may be the best candidates for this form of treatment, since much higher radiation doses are likely to be delivered to these tumor sites, compared to large bulky tumors.

CONCLUSION

This study considered the clinical situation of small tumor targets as the most favorable for efficient RAIT. The dose given was related to the anticipated red marrow dose, and efficacy and toxicity were determined. Favorable tumor targeting, although not perfect, was seen, and the therapy resulted in disease stabilization in some of the patients. The efficacy of RAIT, however, was modest despite the small tumor targets present. Although there is still a need for a definitive Phase II trial to determine the efficacy of RAIT at the MTD, the current results suggest that future dose-intensification trials, including those utilizing stem-cell rescue, should be considered in these patients.

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